Inline SU Viscometer as Process Analytical Technology to Measure Real-time Protein Concentration in TFF Operation

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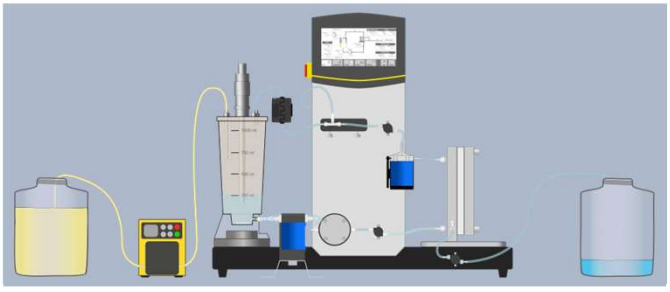
Introduction

TFF is typically performed to formulate biologic therapeutics to high protein concentrations and includes ultrafiltration (UF) as well as diafiltration (DF) steps. During the process, protein concentrations are typically measured by at-line analytics to determine if target protein concentrations are reached to initiate the next processing step. Strongly increasing viscosities at target protein concentrations greater than 100 g/L pose however challenges to the sample measurement process with the risk of inaccurate concentration determination.

Experimental Approach

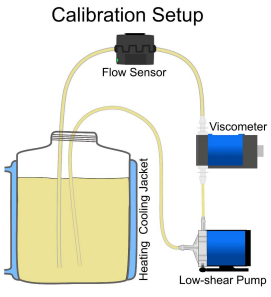
A novel single-use inline viscometer, based on magnetic levitation, was developed to expand the currently available inline process analytical technology (PAT) toolbox. The inline viscometer can be readily integrated into the feed stream of the TFF device. In a first step, a correlation between viscosity, temperature and protein concentration was established by ramping the temperature at various protein concentrations. The resulting polynomial quantification model was then used to evaluate the performance of the inline viscometer to measure protein concentration in a TFF process consisting of two UF and one DF step.



Results and Discussion

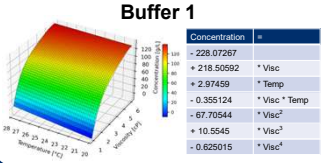
Calibration of Quantification Model:

(a) Protein (mAb) in buffer was recirculated in a calibration setup including a heating and cooling device, starting at the highest protein concentration (140 g/L).
(b) Temperature was ramped from 20-28°C, while measuring the resulting viscosity for each temp-conc combination.
(c) Protein was diluted to the next lower concentration and steps (a) – (c) were repeated until the lowest calibration concentration (0 g/L) was reached.



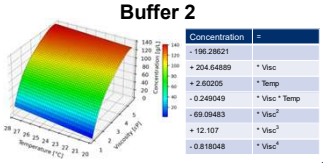
- A reliable correlation between protein concentration and viscosity could be established for each buffer by linear regression, resulting in a polynomial quantification formula.

Buffer 1



Concentration	=
- 228.07267	
+ 218.50592	* Visc
+ 2.97459	* Temp
- 0.355124	* Visc * Temp
- 67.70544	* Visc^2
+ 10.5545	* Visc^3
- 0.625015	* Visc^4

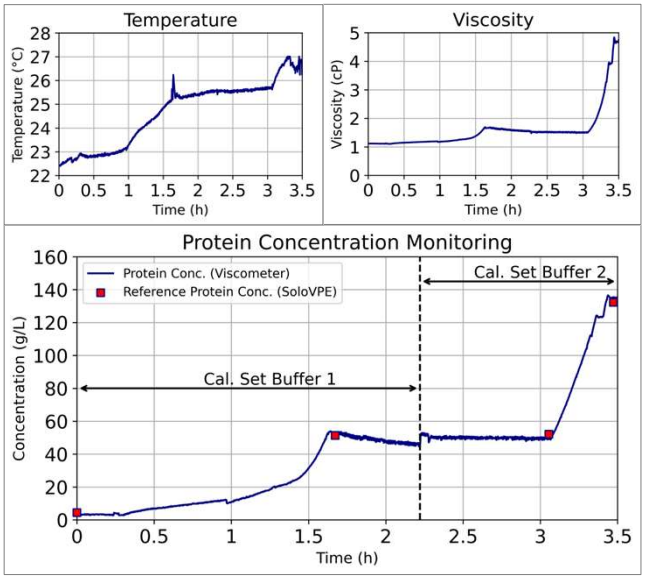
Buffer 2



Concentration	=
- 196.28621	
+ 204.64889	* Visc
+ 2.60205	* Temp
- 0.249049	* Visc * Temp
- 69.09483	* Visc^2
+ 12.107	* Visc^3
- 0.818048	* Visc^4

Process Monitoring:

- Subsequently, real-time monitoring of protein concentration during TFF operation was possible due to real-time temperature and viscosity measurements by the inline viscometer. Concentration could be monitored based on the established polynomial quantification formulas.
- Viscometer based protein quantification aligned well with offline reference analytics.
- Real-time protein concentration measurements enabled accurate process control during the UF and DF steps, without sampling during the process.



Product Quality:

- The implementation of the viscometer had no impact on mAb product quality such as monomer content, low molecular weight species (LMW) and high molecular weight species (HMW).

Conclusion and Outlook

Overall, this study affirms the effectiveness of a novel single-use viscometer in monitoring protein concentration during pharmaceutical processes, making it an ideal inline PAT tool. The real-time viscosity measurement capability has the potential to increase the control of TFF operations and improve process performance.

Acknowledgements

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